

AMENDMENTS TO THE CLAIMS:

This listing of claims will replace all prior versions, and listings, of claims in the application:

LISTING OF CLAIMS:

1-273. Canceled

274. (New) A method for producing and isolating a polypeptide having at least one desirable property comprised of the steps of:

(a) subjecting a starting or parental polynucleotide set to a mutagenesis process so as to produce a progeny polynucleotide set; said mutagenesis process comprising subjecting a codon-containing template polynucleotide to a polymerase-based amplification using a plurality of degenerate oligonucleotides for each codon to be mutagenized, where each of said degenerate oligonucleotides contains a degenerate triplet sequence, so as to generate a plurality of progeny polynucleotides, and

(b) subjecting the progeny polynucleotide set to an end selection-based screening and enrichment process, so as to select for a desirable subset of the progeny polynucleotide set;

whereby the above steps can be performed iteratively and in any order, combination, and permutation,

whereby the end selection-based process of step (b) creates ligation-compatible ends,

whereby the creation of ligation-compatible ends in step (b) is optionally used to facilitate one or more intermolecular ligations, that are preferably directional ligations, within members of the progeny polynucleotide set so as to achieve assembly and/or reassembly mutagenesis,

whereby the creation of ligation-compatible ends in step (b) serves to facilitate ligation of the progeny polynucleotide set into an expression vector system and expression cloning,

whereby the expression cloning of the progeny polynucleotide set serves to generate a polypeptide set,

whereby the generated polypeptide set can be subjected to an expression screening process, and

whereby expression screening of the progeny polypeptide set provides a means to identify a desirable species.

275. (New) The method of claim 274, wherein said degenerate oligonucleotide comprises a degenerate triplet sequence for each codon to be mutagenized.

276. (New) The method of claim 274, wherein said degenerate oligonucleotide comprises a 32-fold degenerate triplet sequence for each codon to be mutagenized.

277. (New) The method of claim 276, wherein said 32-fold degenerate triplet sequence is selected from the group consisting of nng/t, nnc/g, and nna/c.

278. (New) The method of claim 274, wherein said end selection-based process comprises a selection marker containing a topoisomerase recognition site.

279. (New) The method of claim 278, wherein said topoisomerase recognition site comprises a topoisomerase I recognition site.

280. (New) The method of claim 279, wherein said topoisomerase I recognition site comprises a vaccinia topoisomerase recognition site.

281. (New) The method of claim 274, wherein said end selection-based process comprises a selection marker containing a restriction enzyme recognition site selected from the group consisting of BamHI, BglII, PstI, XbaI, AscI, NotI, PacI, PmeI, SrfI, Sse838I, SwaI, BspGI, Eco57I, and any combination thereof.

282. (New) A method for producing and isolating a polypeptide having at least one desirable property comprising:

(a) subjecting a starting or parental polynucleotide set to a mutagenesis process so as to produce a progeny polynucleotide set; said mutagenesis process comprising in vitro shuffling, in vivo shuffling or a combination of in vitro and in vivo shuffling; and

(b) subjecting the progeny polynucleotide set to an end selection-based screening and enrichment process, so as to select for a desirable subset of the progeny polynucleotide set;

whereby the above steps can be performed iteratively and in any order, combination, and permutation,

whereby the end selection-based process of step (b) creates ligation-compatible ends,

whereby the creation of ligation-compatible ends in step (b) is optionally used to facilitate one or more intermolecular ligations, that are preferably directional ligations, within members of the progeny polynucleotide set so as to achieve assembly and/or reassembly mutagenesis,

whereby the creation of ligation-compatible ends in step (b) serves to facilitate ligation of the progeny polynucleotide set into an expression vector system and expression cloning,

whereby the expression cloning of the progeny polynucleotide set serves to generate a polypeptide set,

whereby the generated polypeptide set can be subjected to an expression screening process, and

whereby expression screening of the progeny polypeptide set provides a means to identify a desirable species.

283. (New) The method of claim 282, wherein said end selection-based process comprises a selection marker containing a topoisomerase recognition site.

284. (New) The method of claim 283, wherein said topoisomerase recognition site comprises a topoisomerase I recognition site.

285. (New) The method of claim 284, wherein said topoisomerase I recognition site comprises a vaccinia topoisomerase recognition site.

286. (New) The method of claim 282, wherein said end selection-based process comprises a selection marker containing a restriction enzyme recognition site selected from the group consisting of BamHI, BglII, PstI, XbaI, AscI, NotI, PacI, PmeI, SrfI, Sse838I, SwaI, BspGI, Eco57I, and any combination thereof.